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FILE 'MEDLINE' ENTERED AT 11:14:02 ON 16 JUL 2003

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=> s (atp sulfurylase and aps kinase) and dna
L1          52 (ATP SULFURYLASE AND APS KINASE) AND DNA

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PROCESSING COMPLETED FOR L1
L2          29 DUP REM L1 (23 DUPLICATES REMOVED)

=> s l2 and (human ir mouse)
L3           0 L2 AND (HUMAN IR MOUSE)

=> s l2 and (human or mouse)
L4           9 L2 AND (HUMAN OR MOUSE)

=> s l4 and 1990-1999/py
L5           6 L4 AND 1990-1999/PY

=> d l5 1-6 ibib ab

L5   ANSWER 1 OF 6     MEDLINE
ACCESSION NUMBER:   2000026854     MEDLINE
DOCUMENT NUMBER:    20026854   PubMed ID: 10559207
TITLE:              Genomic organization of the mouse and
                    human genes encoding the ATP
                    sulfurylase/adenosine 5'-phosphosulfate kinase
                    isoform SK2.
AUTHOR:             Kurima K; Singh B; Schwartz N B
CORPORATE SOURCE:   Department of Pediatrics, University of Chicago, Chicago,
                    Illinois 60637, USA.
CONTRACT NUMBER:    AR-19622 (NIAMS)
     HD-17332 (NICHD)
SOURCE:             JOURNAL OF BIOLOGICAL CHEMISTRY, (1999 Nov 19)
                    274 (47) 33306-12.
                    Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY:       United States
DOCUMENT TYPE:      Journal; Article; (JOURNAL ARTICLE)
LANGUAGE:           English
FILE SEGMENT:       Priority Journals
OTHER SOURCE:       GENBANK-AF172857; GENBANK-AF172858; GENBANK-AF172859;
                    GENBANK-AF172860; GENBANK-AF172861; GENBANK-AF172862;
                    GENBANK-AF172863; GENBANK-AF172864; GENBANK-AF172865;
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                    GENBANK-AF173363; GENBANK-AF173364; GENBANK-AF173365
ENTRY MONTH:        199912
ENTRY DATE:         Entered STN: 20000113
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Last Updated on STN: 20000113
Entered Medline: 19991214

AB Mammalian **ATP sulfurylase**/adenosine 5'-phosphosulfate (**APS**) **kinase** consists of kinase and sulfurylase domains, and catalyzes two sequential reactions to synthesize the universal sulfate donor, phosphoadenosine phosphosulfate (PAPS). In simpler organisms, the **ATP sulfurylase** and **APS kinase** reactions are catalyzed by separate enzymes encoded by two or three genes, suggesting that a fusion of separate genes during the course of evolution generated the bifunctional enzyme. We have characterized the genomic structure of the PAPS synthetase SK2 isoform genes for **mouse** (MSK2) and **human** (HSK2) and analyzed the possible fusion region. The MSK2 and HSK2 genes exhibit a common structure of 13 exons, including a 15-nucleotide alternatively spliced exon 8. Enzyme activities of several bacterially expressed exon assemblages showed exons 1-6 encode **APS kinase**, while exons 6-13 encode **ATP sulfurylase**. The MSK2 construct without the exon 6-encoded peptide showed no kinase or sulfurylase activity, demonstrating that exon 6 encodes sequences required for both activities. Exon 1 and its 5'-flanking sequence are highly divergent between the two species, and intron 1 of the HSK2 gene contains a region similar to the MSK2 promoter sequence, suggesting that it may be the remnant of a now-superceded regulatory region. The HSK2 promoter contains a GC-rich region, not present in the **mouse** promoter, and has few transcription factor binding sites in common with MSK2. These differences in the two promoter regions suggest that species-specific mechanisms regulate expression of the SK2 isoform.

L5 ANSWER 2 OF 6 MEDLINE
ACCESSION NUMBER: 1998312048 MEDLINE
DOCUMENT NUMBER: 98312048 PubMed ID: 9648242
TITLE: cDNA cloning, expression, and characterization of the **human** bifunctional **ATP sulfurylase**/adenosine 5'-phosphosulfate kinase enzyme.
AUTHOR: Yanagisawa K; Sakakibara Y; Suiko M; Takami Y; Nakayama T; Nakajima H; Takayanagi K; Natori Y; Liu M C
CORPORATE SOURCE: Department of Biochemistry, University of Texas Health Center, Tyler 75710, USA.
SOURCE: BIOSCIENCE, BIOTECHNOLOGY, AND BIOCHEMISTRY, **(1998 May)** 62 (5) 1037-40.
Journal code: 9205717. ISSN: 0916-8451.
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AF033026
ENTRY MONTH: 199807
ENTRY DATE: Entered STN: 19980811
Last Updated on STN: 20000303
Entered Medline: 19980730

AB A cDNA encoding the **human** bifunctional **ATP sulfurylase**/adenosine 5'-phosphosulfate (**APS**) **kinase** was cloned and sequenced. The enzyme contains an **APS kinase** domain in its N-terminal portion and an **ATP sulfurylase** domain in its C-terminal portion. Recombinant full-length enzyme and its constituent **APS kinase** and **ATP sulfurylase** domains were individually expressed, purified, and shown to have their respective enzymatic activities.

L5 ANSWER 3 OF 6 MEDLINE
ACCESSION NUMBER: 96094345 MEDLINE
DOCUMENT NUMBER: 96094345 PubMed ID: 7493984
TITLE: The isolation and characterization of cDNA encoding the

mouse bifunctional **ATP sulfurylase**-adenosine 5'-phosphosulfate kinase.

AUTHOR: Li H; Deyrup A; Mensch J R Jr; Domowicz M; Konstantinidis A K; Schwartz N B
CORPORATE SOURCE: Department of Pediatrics, University of Chicago, Illinois 60637, USA.
CONTRACT NUMBER: AR-19622 (NIAMS)
HD-09402 (NICHD)
HD-17332 (NICHD)
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, **(1995 Dec 8)** 270 (49) 29453-9.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-L39001; GENBANK-M68858; GENBANK-M74586; GENBANK-M94886; GENBANK-S55315; GENBANK-T09181; GENBANK-U05218; GENBANK-U05238; GENBANK-U07353; GENBANK-U34883; GENBANK-X60157; GENBANK-X79053
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AB Biosynthesis of the activated sulfate donor, adenosine 3'-phosphate 5'-phosphosulfate, involves the sequential action of two enzyme activities: **ATP sulfurylase**, which catalyzes the formation of adenosine 5'-phosphosulfate (APS) from ATP and free sulfate, and **APS kinase**, which subsequently phosphorylates APS to produce adenosine 3'-phosphate 5'-phosphosulfate. Oligonucleotide primers were derived from a **human** infant brain-expressed sequence tag putatively encoding a portion of **APS kinase**. Using these primers, reverse transcriptase-polymerase chain reaction was performed on mRNA from neonatal normal mice resulting in amplification of a 127-bp **DNA** fragment. This fragment was subsequently used to screen a **mouse** brain lambda gt11 cDNA library, yielding a 2.2-kb clone. Primers were designed from the 5'-end of the 2.2-kb clone, and 5'-rapid amplification of cDNA ends was used to obtain the translation start site. Sequence from the overlapping clones was assembled into a 2475-bp composite sequence, which contains a single open reading frame that translates into a 624-deduced amino acid sequence. Northern blots of total RNA from neonatal mice yielded a single message species at approximately 3.3 kb. Southern blot of genomic **DNA** digested with several restriction enzymes suggested the gene is present as a single copy. Comparison against sequence data bases suggested the composite sequence was a fused sulfurylase-kinase product, since the deduced amino acid sequence showed extensive homology to known separate sequences of both **ATP sulfurylase** and **APS kinase** from several sources. The first 199 amino acids corresponded to **APS kinase** sequence, followed by 37 distinct amino acids, which did not match any known sequence, followed by 388 amino acids that are highly homologous to known **ATP sulfurylase** sequences. Finally, recombinant enzyme expressed in COS-1 cells exhibited both **ATP sulfurylase** and **APS kinase** activity.

L5 ANSWER 4 OF 6 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 1999:526237 BIOSIS
DOCUMENT NUMBER: PREV199900526237
TITLE: Chemical modification and site-directed mutagenesis of conserved HXXH and PP-loop motif arginines and histidines in the murine bifunctional **ATP sulfurylase**/adenosine 5'-phosphosulfate kinase.
AUTHOR(S): Deyrup, Andrea T.; Singh, Bhawani; Krishnan, Srinivasan; Lyle, Stephen; Schwartz, Nancy B. (1)

CORPORATE SOURCE: (1) Dept. of Pediatrics, University of Chicago, 5841 S. Maryland Ave., Chicago, IL, 60637 USA
SOURCE: Journal of Biological Chemistry, (**Oct. 8, 1999**) Vol. 274, No. 41, pp. 28929-28936. ISSN: 0021-9258.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The sulfurylase domain of the **mouse** bifunctional enzyme **ATP sulfurylase**/adenosine 5'-phosphosulfate (**APS**) **kinase** contains HXXH and PP-loop motifs. To elucidate the functional importance of these motifs and of conserved arginines and histidines, chemical modification and site-directed mutagenesis studies were performed. Chemical modification of arginines and histidines with phenylglyoxal and diethyl pyrocarbonate, respectively, renders the enzyme inactive in sulfurylase, kinase, and overall assays. Data base searches and sequence comparison of bifunctional **ATP sulfurylase /APS kinase** and monofunctional ATP sulfurylases shows a limited number of highly conserved arginines and histidines within the sulfurylase domain. Of these conserved residues, His-425, His-428, and Arg-421 are present within or near the HXXH motif whereas His-506, Arg-510, and Arg-522 residues are present in and around the PP-loop. The functional role of these conserved residues was further studied by site-directed mutagenesis. In the HXXH motif, none of the alanine mutants (H425A, H428A, and R421A) had sulfurylase or overall activity, whereas they all exhibited normal kinase activity. A slight improvement in reverse sulfurylase activity (< 10% residual activity) and complete restoration of forward sulfurylase was observed with R421K. Mutants designed to probe the PP-loop requirements included H506A, R510A, R522A, R522K, and D523A. Of these, R510A exhibited normal sulfurylase and kinase activity, R522A and R522K showed no sulfurylase activity, and H506A had normal sulfurylase activity but produced an effect on kinase activity (< 10% residual activity). The single aspartate, D523A, which is part of the highly conserved GRD sequence of the PP-loop, affected both sulfurylase and kinase activity. This mutational analysis indicates that the HXXH motif plays a role only in the sulfurylase activity, whereas the PP-loop is involved in both sulfurylase and kinase activities. Residues specific for sulfurylase activity have also been distinguished from those involved in kinase activity.

L5 ANSWER 5 OF 6 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 1998:390189 BIOSIS
DOCUMENT NUMBER: PREV199800390189
TITLE: Molecular cloning, expression, and characterization of **human** bifunctional 3'-phosphoadenosine 5'-phosphosulfate synthase and its functional domains.
AUTHOR(S): Venkatachalam, K. V.; Akita, Harukuni; Strott, Charles A. (1)
CORPORATE SOURCE: (1) NICHD, Build. 49, Rm. 6A36, National Institutes Health, Bethesda, MD 20892-4510 USA
SOURCE: Journal of Biological Chemistry, (**July 24, 1998**) Vol. 273, No. 30, pp. 19311-19320. ISSN: 0021-9258.
DOCUMENT TYPE: Article
LANGUAGE: English

AB The universal sulfonate donor, 3'-phosphoadenosine 5'-phosphosulfate (PAPS), is synthesized by the concerted action of **ATP sulfurylase** and adenosine 5'-phosphosulfate (**APS**) **kinase**, which in animals are fused into a bifunctional protein. The cDNA for **human** PAPs synthase (hPAPSS) along with polymerase chain reaction products corresponding to several NH2- and COOH-terminal fragments were cloned and expressed in COS-1 cells. A 1-268-amino acid fragment expressed **APS kinase** activity, whereas a 220-623 fragment evinced **ATP sulfurylase** activity. The 1-268 fragment and full-length hPAPSS (1-623) exhibited hyperbolic

responses against APS substrate with equivalent Km values (0.6 and 0.4 muM, respectively). The 1-268 fragment demonstrated Michaelis-Menten kinetics against ATP as substrate (Km 0.26 mM); however, full-length hPAPSS exhibited a sigmoidal response (apparent Km 1.5 mM) suggesting cooperative binding. Catalytic efficiency (Vmax/Km) of the 1-268 fragment was 64-fold higher than full-length hPAPSS for ATP. The kinetic data suggest that the COOH-terminal domain of hPAPSS exerts a regulatory role over **APS kinase** activity located in the NH2-terminal domain of this bifunctional protein. In addition, the 1-268 fragment and full-length hPAPSS were overexpressed in Escherichia coli and column purified. Purified full-length hPAPSS, in contrast to the COS-1 cell-expressed cDNA construct, exhibited a hyperbolic response curve against ATP suggesting that hPAPSS is perhaps modified in vivo.

L5 ANSWER 6 OF 6 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 1999-12918 BIOTECHDS
TITLE: **Human**-derived **APS-kinase/ATP-sulfurylase** gene; **human** recombinant **APS-kinase -ATP-sulfurylase** production in Escherichia coli, useful for the large-scale production of phosphoadenosine-phosphosulfate
PATENT ASSIGNEE: ZH-Human-Sci.Shinko-Zaid; Untika
LOCATION: Japan.
PATENT INFO: JP 11187883 **13 Jul 1999**
APPLICATION INFO: JP 1997-360387 26 Dec 1997
PRIORITY INFO: JP 1997-360387 26 Dec 1997
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
OTHER SOURCE: WPI: 1999-451549 [38]
AB A **human**-derived **APS-kinase/ATP-sulfurylase** (I) and its encoding **DNA** sequence (II) is claimed. Also claimed are: variants of (I) and (II); a vector and host cells, e.g. Escherichia coli, containing (II); the recombinant preparation of (I); and a method for the production of 3 -phosphoadenosine-5 -phosphosulfate (PAPS) using the recombinant (I). The invention can prepare PAPS in large amounts. In an example, (I) was identified and expressed in E. coli DE3 and PAPS produced. (9pp)

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FULL ESTIMATED COST	31.59	31.80

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RA   Superti-Furga A., Haque S., Abbas H., Ahmad W., Ahmad M., Cohn D.H.;
RT   "Mutations in orthologous genes in human spondyloepimetaphyseal dysplasia
RT   and the brachymorphic mouse";
RL   Nat. Genet. 20(2):157-162(1998).
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RN   [2]
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RA   Swank R.T., Superti-Furga A., Haque S., Abbas H., Ahmad W., Ahmad M.,
RA   Cohn D.H.;
RT   ;
RL   Submitted (08-SEP-1998) to the EMBL/GenBank/DDBJ databases.
RL   Pediatrics, Medical Genetics, Cedars-Sinai Research Institute, 8700 Beverly
RL   Blvd., Los Angeles, CA 90048, USA
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Saidha, T kchand

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1. J. Biol. Chem. (1999, Nov 19), 274 (47), 33306-12.
2. J. Biol. Chem. (1999, April 16), 274 (16), 10751-57
3. International J. of Biochem. & Cell Biology (May 1999), 31 (5) : 613-26.
4. FASEB Journal (May 1998), 12 (7) : 603-12.
5. Gene (Nov 20, 1995) 165 (2), 243-8.

Thank you

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